Desalination and Water Treatment
www.deswater.com

doi: 10.5004/dwt.2023.29145

281 (2023) 58–69
January

Enhancement of anaerobic membrane bioreactor performance using microbe activator in palm oil mill effluent treatment

Rou Hui Wong[a], Kai Jun Tan[a], Xin Xin Guo[a], Chin Heng Gan[b], Mohammed J K Bashir[a], Ping Zheng[c], Choon Aun Ng[a,*]

[a]*Department of Environmental Engineering, Engineering and Green Technology Faculty, Universiti Tunku Abdul Rahman, 31900 Kampar, Perak, Malaysia, Tel.: 605-4688888 Ext: 2256; Fax: 605-4667449; emails: ngca@utar.edu.my (C.A. Ng), hui_wong@hotmail.com (R.H. Wong), kaijun1927@gmail.com (K.J. Tan), guoxinxin122@hotmail.com (X.X. Guo), jkbashir@utar.edu.my (M.J.K. Bashir)*
[b]*Department of Civil Engineering, Lee Kong Chian Faculty of Engineering and Science, Universiti Tunku Abdul Rahman, 43000 Cheras, Kajang, Selangor, Malaysia, email: ganch@utar.edu.my*
[c]*State Key Laboratory of Environmental Criteria and Risk Assessment, Chinese Research Academy of Environmental Sciences, Beijing 100012, China, email: zengping71@hotmail.com*

Received 31 December 2021; Accepted 11 November 2022

ABSTRACT

Anaerobic membrane bioreactors (An-MBRs) are a proven technology in wastewater treatment. However, the operation of anaerobic membrane bioreactors (An-MBRs) is inevitably plagued by membrane fouling due to the action of foulants that hinders its effectiveness. Among the foulants, extracellular polymeric substances (EPS) had been identified as the major contributor towards membrane fouling, which suggest that controlling EPS production is crucial towards membrane fouling reduction. Polysaccharide and protein are the two recognized major components of EPS. This explains why in this research, polysaccharide and protein concentrations were used to measure the presence of EPS in the bioreactors. Microbe activator was added into the anaerobic membrane bioreactor (An-MBR) under different dilution factors to assess their efficiencies in promoting the performance of An-MBRs in terms of EPS and membrane fouling control. The An-MBR that was added with 1/500 dilution of microbe activator showed the highest chemical oxygen demand removal efficiency which is 79.47% ± 2.76%, while the other 3 An-MBRs with 1/125 dilution, 1/1,000 dilution and without added with microbe activator had removal efficiencies of 55.09% ± 4.25%, 56.42% ± 3.36% and 53.49% ± 4.09%, respectively. The bioreactors added with microbe activator with the dilution of 1/500 also achieved the highest EPS removal efficiency mainly in terms of polysaccharides. Also, it had the lowest membrane fouling rate during membrane filtration among the An-MBRs. It is speculated that the introduction of suitable doses of microbe activator would contribute a suitable amount of light metal cations into the bioreactor. Right concentration of light metal cations could enhance microbial activities which may help to control EPS productions. For other dilution factors, the performance of An-MBR was not enhanced. This may be due to the excessive or too little concentration of microbe activator would not improve the performance of An-MBR.

Keywords: Anaerobic membrane bioreactor; Extracellular polymeric substances; Membrane fouling; Microbe activator; Palm oil mill effluent

* Corresponding author.

Presented at the 4th International Symposium on Green and Sustainable Technology 2021 (ISGST 2021), 3–6 October 2021, Ipoh, Malaysia

1. Introduction

Increasing demand for clean water as well as concerns from the public, had directly and indirectly urged the wastewater treatment plant (WWTP) operators to enhance their effluent quality. As such, several new technologies were developed in terms of physical, biological, or chemical processes. Most notably, the membrane bioreactor (MBRs) processes which combine activated sludge process to that of the membrane filtration process. MBRs processes have been proven as an effective means in wastewater treatment and are being widely adopted in various countries namely China, USA, EU etc. Zanetti et al. [1] stated that MBRs are space savings, eliminate the needs of additional biological treatment units and fewer sludge production when compared to conventional activated sludge treatment.

In general, MBRs technologies can be summarized into two main conditions, which are being termed as aerobic MBRs and anaerobic MBRs. Their operating mode is the main component that differentiates them. Aerobic MBRs require continuous aeration to supply oxygen to the microbial in the sludge flocs [2]. Therefore, the drawbacks are high energy consumption [3] and thus higher cost to operate aerobic MBRs. By comparison, anaerobic MBRs do not require aeration in the digestion process, while simultaneously it has the potential to perform energy recovery from the methane gas produced [4]. Other than the above-mentioned drawbacks of aerobic MBRs, the high concentration of organic contaminants within palm oil mill effluent (POME) and its high temperature properties shows that anaerobic MBR is a more suitable treatment method [5]. There are many methods used for POME treatment as stated in Table 1. From Table 1, it shows that up-flow sludge blanket (UASB) bioreactor in POME treatment had the best performance in chemical oxygen demand (COD) removal efficiency up to 96%.

There is one common problem in the operation of either aerobic MBRs or anaerobic MBRs – the membrane fouling issues. Membrane fouling will cause membranes to age faster and increase the operational cost of MBRs from the cleaning stages.

Extracellular polymeric substances (EPS) that is in great interest of research nowadays mainly due to its dominant role in the fouling processes. EPS consists of protein, polysaccharides and humic acids [4]. It is reported that EPS directly influences the properties of sludge flocs in terms of their hydrophobicity, adhesion, flocculation, settling and dewatering.

EPS production plays a vital part towards membrane fouling control. It is regulated by different types of factors. According to Wu and Fane [6], unsteady organic loading rate (OLR) may accelerate polysaccharides production, which ultimately increases membrane fouling. Lin et al. [7] had stated that higher levels of cell death and lysis that arose from stress/unfavourable conditions may cause the release of proteins and polysaccharides into the sludge suspension [7], which in turn increased the membrane fouling propensity. Meng et al. [8] found that EPS production control can be achieved by adding enhancer into the sludge flocs or by controlling the feedwater characteristics. Feedwater characteristics are closely related to the metabolism of microorganisms that leads to the EPS production.

Despite that, it is important to note that although EPS serves as an important membrane foulants, a well-optimized EPS concentration needs to present in the treatment plant where the membrane fouling propensity can be minimized. Research by Du et al. [9] revealed that EPS is a key factor towards sludge agglomeration, whereas smaller EPS concentration causes a reduction in sludge agglomeration, which decreases the flocs sizes. As a result, membrane fouling increases as smaller size flocs are more easily deposited on the surface of the membrane.

In real-life applications, chemical methods are often combined with that of the physical cleaning to achieve a better balance in membrane fouling control. The effectiveness of chemical cleaning is well-proven, but it reduces the lifespan of the membrane and may induce secondary pollution. In more recent years, biological ways to control the membrane fouling had gained highlights in the research field due to its sustainability.

Besides utilizing enzymes in biological control, adding vital nutrients into the reactors to aid in the digestion process might improve the outcomes especially in terms of EPS production control. The microbe activator (it is named as TM Agricultural (TM Agri) by the company who produced it) had shed lights on its potential to do so. It is a product to treat algae blooming and had been proven to enhance the yield of farmlands by activating the local soil microbe in inactive or dormant condition. The microbe activator acts as a signal substance that arouses the dormant beneficial microbial, which accelerates the reproduction process and subsequently enhances the microbial community. TM Agriculture (TM Agri) is a liquid formula that is applied directly to the soil and plant. It is acting as a role of soil rejuvenation that helps to promote the breeding of beneficial microbes that have been dormant, helping to improve the microbial activity in the soil. While using in the soil, the core substances of TM Agri will act as signal substances that can arouse the dormant indigenous beneficial microorganisms in the soil in the beginning. The reproduction of microorganisms will be accelerated by the signal substances, thereby rejuvenating the diversity of soil microbial communities and reconstruct a healthy soil micro-ecological system. Then, the composition of TM Agri includes enzymes, amino acids and plants proteins which are extracted from various plants. These plant secretions can provide "first meals" for the revived soil microorganisms to complete their initial reproduction. Finally, a healthy soil micro-ecological system is formed due to the increased number of soil microorganisms. In the same concept, it is believed that it can activate and provide nutrients such as nitrogen and sulphur which are essential for the growth of anaerobic microorganisms [10].

With a higher microbial activity, processes such as nitrogen fixation can be improved. The rate of decomposition on the organic matter can be carried out in an accelerated manner and hence a more productive yield can be achieved. The healthy microbe will produce relatively less EPS and can biodegrade the pollutants better. The similar trend was reported as EPS production is accelerated by

Table 1
Comparison of POME treatment between anaerobic MBRs and non-anaerobic MBRs

No.	Anaerobic MBRs	Non-anaerobic MBRs	References
1	POME treatment by using anaerobic MBR with SRT of 90 d and under thermophilic condition achieved COD removal rate over 98%		[12]
2		Microalgae was used to treat POME. The removal efficiencies were 62.07% for total nitrogen, 47.09% for COD, and 30.77% for total phosphorus	[13]
3		Performance of a laboratory-scale moving bed biofilm reactor (MBBR) and its microbial diversity in palm oil mill effluent (POME) treatment showed the best removal rate of COD (59.4%) and NH_3–N (94.4%)	[14]
4	Synthetic POME was treated with over 98% of COD removal efficiency by using a lab-scale cross-flow anaerobic MBR system		[15]
5	A two-stage submerged anaerobic membrane bioreactor (2-sAn-MBR) was able to achieve COD removal rate up to 70% in POME treatment process		[16]
6		Thermophilic anaerobic digestion performance in POME treatment in terms of COD, BOD, TSS, and O&G removal rate was 80.63% ± 0.46%, 81.01% ± 1.16%, 80.72% ± 0.16%, and 80.02% ± 0.11%, respectively	[17]
7		Up-flow sludge blanket (UASB) bioreactor in POME treatment had COD removal efficiency up to 96%	[18]
8		Natural plant-based fenugreek (*Trigonella foenum-graecum*) coagulant and okra (*Abelmoschus esculentus*) flocculant for palm oil mill effluent (POME) treatment had pollutant removal efficiencies of 94.97%, 92.70% and 63.11% in turbidity, TSS and COD, respectively	[19]
9		Performance of photocatalytic fuel cells (PFCs) in the treatment of diluted palm oil mill effluent (POME) in COD removal rate is 74%	[20]
10		Performance of ultrasonic-assisted membrane anaerobic system in palm oil mill effluent (POME) treatment had COD removal efficiencies of 82.75% and 94.43% for the final discharge pond and decanter processing unit samples respectively	[21]
11		Palm kernel shell-based adsorbent in treating POME had removal rate of 99.7% of the initial color and 85.0% of the initial COD	[22]

unfavourable conditions or environmental stresses as a direct response of bacteria to protect themselves [7–11].

The product had never been applied in the wastewater treatment process, especially those involving high strength wastewater such as POME. From the above context, it is shown that the EPS production is mainly controlled by the microbial activities [11]. Hence, it is believed that by adding suitable concentration of the microbe activator into the anaerobic MBRs, it improves the digestion process by activating those beneficial microbes from a dormant state that could potentially results in an improved biological outcome which reduces the EPS production in the digester.

The performance of the MBR in treating wastewater is varied due to different operation parameters such as mixed liquor volatile suspended solids (MLVSS), mixed liquor suspended solids (MLSS), sludge retention time (SRT), hydraulic retention time (HRT), pH, microbial floc size, temperature, type of effluent and its concentration etc. This project is focused mainly on microbial activator (TM Agri) to test its efficiency in enhancing the performance

of MBR in treating POME. The same operation parameters were applied to all the anaerobic membrane bioreactors (An-MBRs) involved in this project. It may not be fair to compare the results from this project's finding with the performance of other reported MBRs as the operating parameters were not optimized.

2. Methods

2.1. Collection of POME

POME was collected from Tian Siang Palm Oil Mill located in Air Kuning, Perak, Malaysia. The POME was filtered by a sieve with mesh size of 0.053 mm (No. 270) before feeding it into the bioreactors to remove suspended solids with larger particle size. The characteristic of POME is shown in Table 2.

2.2. Microbe activator

The microbe activator used in this experiment is named TM Agricultural (TM Agri) that is produced by the Best Environmental Technologies Inc., Canada. It is mainly used in agricultural application where the soil and crops performances can be enhanced through the limitation of inorganic material leakage and stimulating strains of beneficial microbes that have been dormant (existing in the soil), thus helping to increase and enhance the microbial activity in the soil. The initial raw concentration of TM Agri is shown in Table 3. In addition, Table 3 also shows the concentrations of different dilution ratios (1:125, 1:500 and 1:1,000) of TM Agri in the 800 mL bioreactors.

2.3. Operation of lab-scale anaerobic bioreactors

The lab-scale anaerobic membrane bioreactors (An-MBRs) were separated into two stages which are anaerobic bioreactor and in-house flat sheet membrane filtration system. Anaerobic bioreactor was set up using 1 L Büchner flask (working volume 800 mL) with a rubber cap with openings for de-sludging, nitrogen gas pumping and sampling purposes. The flask's sidearm was connected to a biogas probe as shown in Fig. 1. In total, 4 sets of anaerobic bioreactors were set up with the following conditions: 1st set is without microbe activator (bioreactor A), 2nd set was added with 1/125 dilution ratio of microbe activator (bioreactor B), 3rd set was added with 1/500 dilution ratio of microbe activator (bioreactor C), 4th set was added with 1/1,000 dilution ratio of microbe activator (bioreactor D). The SRT and HRT of the four bioreactors were fixed at 30 and 12 d, respectively. By fixing the SRT and HRT, the total amount of discharge of anaerobic bioreactors were calculated by Eq. (1) and the amount of sludge and supernatant are calculated by Eqs. (2) and (3), respectively. Table 4 shows a summary of the operating parameters for anaerobic bioreactors.

$$\text{HRT} = \frac{\left(\text{Total Operating Volume of MBR, mL}\right)}{\left(\text{Volume of total feeding per day, mL/day}\right)} \tag{1}$$

$$\text{SRT} = \frac{\left(\text{Total Operating Volume of MBR, mL}\right)}{\left(\text{Volume of desludge per day, mL/day}\right)} \tag{2}$$

$$\text{Volume of supernatant per day}\left(\text{mL/day}\right) = \frac{\text{Volume of total feeding per day}\left(\text{mL/day}\right)}{\text{Volume of desludge per day}\left(\text{mL/day}\right)} \tag{3}$$

2.4. In-house flat sheet membrane

In-house fabricated flat sheet membrane was used in this experiment. The overall membrane fabrication process is as follows: The technique used in membrane casting is the dry/wet phase. The membrane was produced using the semi-auto membrane casting machine. The polymer of choice was polyethersulfone (PES) while simultaneously the solvents, N-methyl-2-pyrrolidone (NMP) solutions were used. To fabricate the membrane, the polymers and the NMP solutions were prepared as dope solutions and then it was slowly added onto the smooth glass plate on top of the membrane casting machine. The knife gap was determined as 10 μm thick, and it serves as the membrane thickness. The membrane characteristics are shown in Table 5.

Table 2
Characterization of POME [23]

Parameters	Mean	Range
pH	4.2	3.4–5.2
Biological oxygen demand (BOD)	25,000	10,250–43,750
Chemical oxygen demand (COD)	51,000	15,000–100,000
Total solids	40,000	11,500–79,000
Suspended solids	18,000	5,000–54,000
Volatile solids	34,000	9,000–72,000
Oil and grease	6,000	130–18,000
Ammoniacal nitrogen	35	4–80
Total nitrogen	750	180–1,400

Note: All the values are in the unit of mg/L except pH.

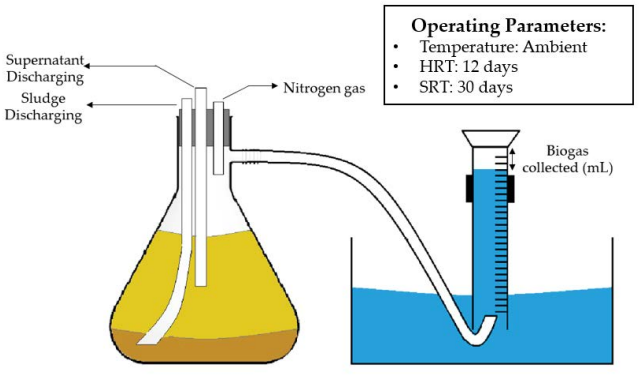

Fig. 1. Schematic diagram of anaerobic bioreactors.

Table 3
Composition of TM Agricultural

Compositions	Initial concentration (mg/L)	Concentration with dilution 1: 125 (mg/L) in the 800 mL bioreactor	Concentration with dilution 1: 500 (mg/L) in the 800 mL bioreactor	Concentration with dilution 1: 1,000 (mg/L) in the 800 mL bioreactor
Nitrogen	61.4	0.55	0.14	0.07
Phosphorus	43.8	0.39	0.10	0.05
Potassium	1,280.0	11.38	2.84	1.42
Calcium	73.0	0.65	0.16	0.08
Magnesium	51.7	0.46	0.11	0.06
Sodium	273.0	2.43	0.61	0.30
Sulfur	186.0	1.65	0.41	0.21
Unlisted trace organic materials	2,184.1	19.41	4.85	2.43
Total feed in concentration in the bioreactor	4,153.0	36.92	9.23	4.61

Table 4
Operating parameters for anaerobic bioreactors

Parameters	A	B	C	D
Temperature, °C	Ambient			
SRT, d	30			
HRT, d	12			
De-sludge per day, mL	40	40	40	40
Supernatant discharge per day, mL	53	53	53	53
Feed in POME, mL	93	93	93	93
Feed in TM Agri ratio	–	1:125	1:500	1:1,000
Feed in microbe activator (TM Agri) concentration, mg/L	–	36.92	9.23	4.61

2.5. Cross-flow filtration process

Fig. 2 shows the schematic diagram of the cross-flow filtration system. The flow rate of the filtration system was set at 200 mL/min and the pressure was fixed. The supernatant collected from the anaerobic bioreactor was added into the water tank and the process was started. The permeate solution was collected for the analytical test.

2.6. Analytical method

The supernatants obtained from each An-MBR were centrifuged at 5,000 rpm for 10 min, then it was diluted using the appropriate dilution factor. The COD content of the diluted samples were measured by using 5220 D Closed Reflux Colorimetric Standard Method, 21st edition. For EPS concentration, the polysaccharides content is measured by using phenol-sulfuric acid method, while the protein content is measured by using Bradford reagent method. UV-Vis spectrophotometry (Jasco UV-Vis spectrophotometer) was used to determine the concentrations of both the protein and polysaccharides. For the MLSS and MLVSS concentrations, the 21st edition of the standard method was used in the analysis process. Besides, particle size analysis was carried out to determine the sludge flocs sizes in terms of volume and numbers using Mastersizer 2000, Malvern Instruments, UK.

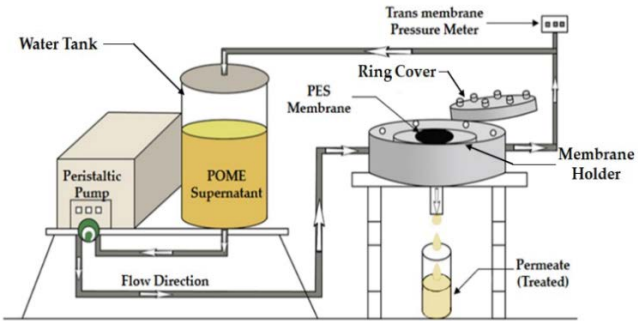

Fig. 2. Schematic diagram of cross-flow filtration process.

Lastly, the membrane fouling control performance of the four An-MBRs was measured by filtering their supernatant through the cross-flow filtration system using in-house flat sheet membranes as per Fig. 2.

3. Results and discussion

3.1. Performances of An-MBRs fed with different dilutions of TM Agri

After feeding the four bioreactors as per Table 4 for three months, their performances in terms of COD removal

Table 5
PES membrane characteristics

Characteristics	Value
Surface area (m^2)	1.46×10^{-3}
Nominal pore size (μm)	0.50
Fixed pressure (bar)	0.10
Pure water flux (L/m^2 h)	80.00

efficiency were analysed and shown in Fig. 3. Among the bioreactors, bioreactor C performed the best by having the highest average COD removal efficiency of 70.60% ± 2.20%, while bioreactor A had the lowest average COD removal efficiency of 34.79% ± 3.69%. The other two bioreactors, namely bioreactors B and D had the average COD removal efficiencies of 38.35% ± 3.64% and 36.91% ± 3.35%, respectively.

Chemical oxygen demand (COD) is one of the single most important criteria in accessing the performances of the An-MBRs. Under fixed operational conditions such as temperature, pH, SRT and HRT, the composition of wastewater and its interactions with the microbial activities had a profound role in the overall COD removal efficiencies. In this study, bioreactor C showed the highest COD removal efficiency, which indicated a better microbe activity in the bioreactor compared to the others.

It took about 2–3 months for the bioreactors to become stable. SRT can be a general guideline in acclimatizing activated sludge (microbe) in the bioreactor. Generally, bioreactors will become stable after being cultivated for a duration of about 2–3 times of SRT. The value of the SRT for all the bioreactors involved in this study is 30 d. This explains why the bioreactors in this study became stable after 2–3 months. Another indication of the stabilization of bioreactors is their MLSS concentration. Stabilized bioreactors have a relatively constant value of MLSS concentration as shown in Fig. 4. Besides that, Fig. 4 also shows that the bioreactor B has the highest MLSS concentration, it indicates that higher concentration of TM Agri can enhance the growth rate of microbial.

According to the material safety data sheet of the microbe activator used [24], the microbe activator formula contains various concentrations of light metal cations including potassium (K^+), sodium (Na^+), calcium (Ca^{2+}) and magnesium (Mg^{2+}). They are being pronounced as able to tie-up excess nutrients through activation of beneficial microbes that limits the growth for algae. In fact, these light metal cations are required nutrients that can improve microbial growth when present in adequate and correct concentration [25]. This is because these metal cations are essentials in enzymatic reaction, inhibitors of sulphide activities, agent-binding nutrients, and biomass stimulants [26]. However, these light metal cations may also cause an inhibitory effect. For example, in the findings reported by Romero-Güiza et al. [27] and Mellyanawaty et al. [28] showed that magnesium ion causes minor and major inhibition of the anaerobic digestion process when present in higher concentration. Milán et al. [29] found that optimum effect can be achieved in anaerobic digestion when their concentrations were adequate (for

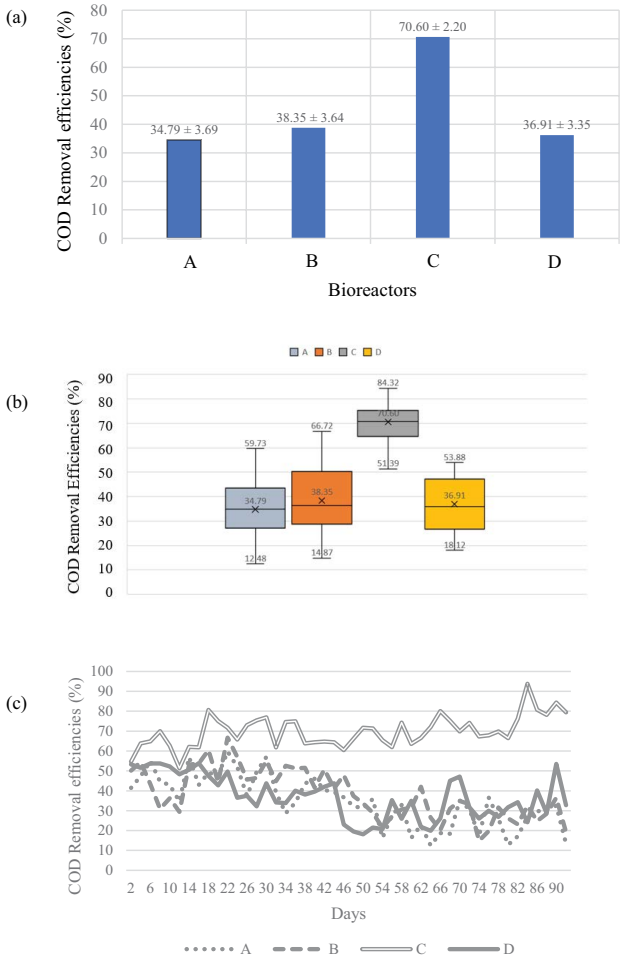

Fig. 3. (a) Average COD removal efficiencies of different bioreactors, (b) Box–plot graph for COD removal efficiencies of different bioreactors and (c) 3 months COD removal efficiencies of different bioreactors in the progressive manner.

examples K^+, Mg^{2+} and Ca^{2+}). Sodium ion has also been reported to induce methane inhibition and poor degradation of organic matter performance when present in high concentration [30]. However, the presence of certain other ions tended to be antagonistic to each other, such as in the case where K^+, Mg^{2+} and Ca^{2+} ions were found to increase the sodium tolerance of anaerobes during anaerobic treatment [31]. Henceforth, an appropriate amount of these metal cations needs to be present so the performances of the bioreactors can be well-balanced and maximized.

From the COD removal result, it can be deduced that 1/500 ratio dilution of the microbe activator in bioreactor C could provide better overall concentration in terms of light metal cations which balances their antagonistic effect and subsequently increases the microbial activities. This in turn helped in the reduction of the COD concentration. As for bioreactors A, B and D, the results are highly similar in terms of COD removal efficiency, this suggests that too high or too low of microbe activator concentration may not have a good effect on the microbial activities.

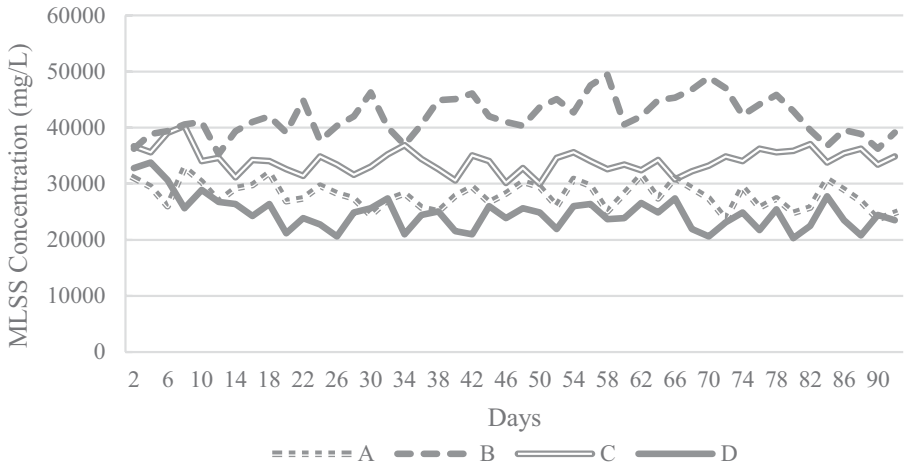

Fig. 4. MLSS concentration throughout 3 months period.

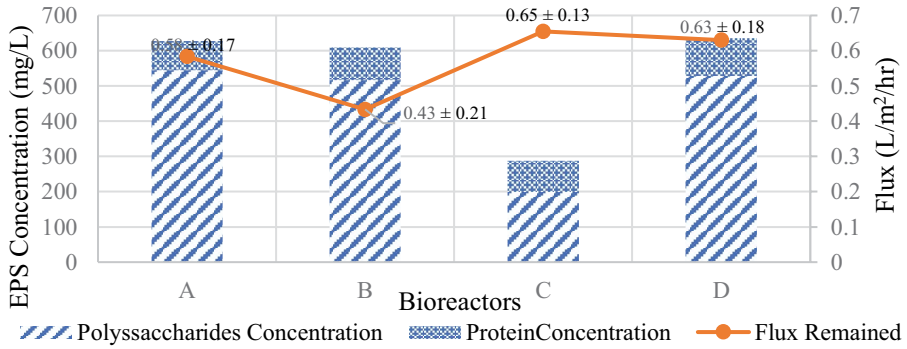

Fig. 5. Relationship between membrane fouling rate and different EPS concentrations.

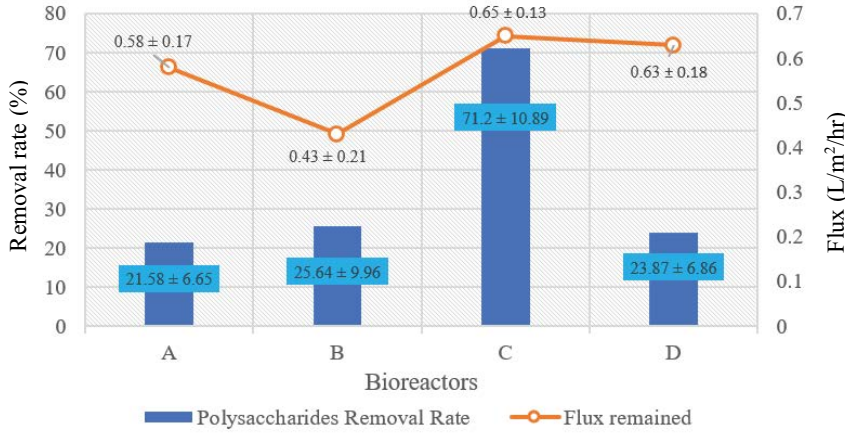

Fig. 6. Relationship between different polysaccharides concentrations removal rate and membrane fouling rate.

The reduction of anaerobic methanogens activities may also be due to the addition of relatively high concentration of sulfur (S) in the microbe activator. Sulfur undergoes oxidation to become sulphate (SO_4^{2-}). Sulfate can promote inhibitory effects on the methanogens. According to Song et al. [32], high sulphate concentration increases competition over methanogenic bacteria and sulphate reducing bacteria (SRB), induce precipitation of non-alkali metal (thus reducing their availability as micro-nutrient) and promote hydrogen sulphate (H_2S) gas production (destroys

cell membrane). This is also supported by the fact that bioreactor B produced little to no biogas production which suggests a higher inhibitory rate on the methanogenic bacteria. As for bioreactor C, it did produce biogas which indicated a higher methanogenic activity.

Besides sulphur, the inhibition of the methanogenic bacteria may also be a resultant of the anoxic condition induced in the bioreactors instead of strictly anaerobic conditions. Methanogenic bacteria are highly susceptible to even very low oxygen concentration and an anoxic condition will significantly lower their already very slow growth rate [33].

Moreover, it is also suspected that formation of foaming in the bioreactors A, B and D resulted in a lower performance of the bioreactors. Foaming would remove active biomass from the liquid phase in the bioreactor, thereby impacting the anaerobic treatment. It can be caused by incomplete metabolism of influent organic matter. However, foaming issue was not observed in bioreactor C. This indicated that adding a suitable amount of microbe activator into the bioreactor would reduce the foaming problem. Delvigne and Lecomte [34] had stated that protein content released by microbial activities aggravated foam formation. Hence, this helps to explain the low foaming problem of the bioreactor C was partial due to its relatively lower EPS concentration production.

EPS reported in this paper is referred to as the combination of both the protein and polysaccharides concentrations. They were reported to have originated from microorganisms that form microbial aggregates [4]. The membrane fouling control of the An-MBRs was measured by a simple test of flux declination over a fixed time (30 min) under fixed pressure condition. The flux was measured by dividing the 30th minute flux with the 5th min flux. Based on Fig. 5, it can be noticed that bioreactor C had the lowest concentration of polysaccharides in supernatant, which corresponds to the lowest total EPS concentration. EPS production is promoted by unfavourable conditions or environmental stresses as a direct response of bacteria to protect themselves [7–11]. This explains the higher EPS concentration in the bioreactors A, B and D as their performance corresponds to their lower COD removal efficiency, which indicates a stress condition that limits the microorganisms' activity.

Based on the filtration results, bioreactor C with the lowest EPS concentration had the best membrane fouling control performance. The finding is consistent with the theory where the EPS concentration is the main factor in membrane fouling propensity.

Despite that, it must be highlighted that high protein concentration do not necessarily accelerate membrane fouling. Polysaccharides are being considered as the main foulant compared to protein mainly due to its hydrophilic and gelling properties. As a result, this allows them to be easily attached on the membrane surface [35]. This is also consistent with the results shown in Fig. 5. Bioreactor C with the best membrane fouling control also had the lowest polysaccharides concentration as per Figs. 5 and 6

However, the results for the bioreactor B deviated from the expectations. Although bioreactor B had the 2nd lowest EPS concentration, its membrane flux remained after 30 min of filtration showed the lowest among the four, which corresponds to a higher membrane fouling. This may be due to the higher MLSS concentration in the bioreactor B as per Fig. 4.

Table 6 shows that the removal efficiencies of polysaccharides and COD before and after membrane filtration of An-MBRs. The membrane filtration is able to remove EPS effectively regardless of their concentrations in the bioreactors. This is not a case for COD removal. This may be due to pollutants contributed to the COD concentration consisted of dissolved foulants. By comparison, bioreactor C added with the microbe activator in the ratio of 1:500 performed best in the COD removal rate. This indicates that by adding the right microbe activator ratio into the bioreactor is important to produce active microbial which are able to control COD pollutants more effectively. It can be concluded that membrane filtration process can partially treat the POME by (i) stopping the fine particle from going out through the membrane and (ii) the biofilm formed on the membrane surface helps to biodegrade the fine pollutants.

It had been reported by many researchers [36–40] that microbial floc size played an important role in membrane fouling control where membrane fouling increases when the concentration of submicron particles increases. Bioreactor with bigger microbial floc size contributed the least fouling to the membrane. However, in this study, the microbial floc size in all the bioreactors had a quite similar size as per Fig. 7. It indicated that microbe activator (TM Agri) did not affect the size of the floc.

MLSS and MLVSS concentrations in this study were expressed in terms of MLVSS/MLSS ratios and the effect of microbe activator towards their concentration are discussed here. The results are shown in Fig. 8. Based on the results shown, the effect of microbe activator on the MLSS and MLVSS concentrations is inconclusive. The values of MLSS concentrations of the four bioreactors are between 24,700 to 42,400 mg/L throughout this study and attained steady state of microbial activities after three months of cultivation. The parameter is subjected to the growth kinetics of the microbial which can be affected by various factors. It was found in this study, higher MLVSS concentration could

Table 6
Removal efficiencies of polysaccharides and COD before and after membrane filtration of An-MBRs

Parameters	A	B	C	D
Removal efficiencies of polysaccharides BEFORE membrane filtration (%)	21.58 ± 6.65	25.64 ± 9.96	71.20 ± 10.98	23.87 ± 6.86
Removal efficiencies of polysaccharides AFTER membrane filtration (%)	90.89 ± 3.05	90.58 ± 4.46	91.85 ± 3.29	88.96 ± 3.10
Removal efficiencies of COD BEFORE membrane filtration (%)	34.79 ± 3.69	38.35 ± 3.64	70.60 ± 2.22	36.91 ± 3.35
Removal efficiencies of COD AFTER membrane filtration (%)	53.49 ± 3.75	56.42 ± 2.06	79.47 ± 3.74	55.09 ± 5.36

be obtained by adding higher concentrations of TM Agri into the bioreactor. However, having higher MLSS concentration or adding higher concentration of microbe activator is not necessary would improve the performance of the bioreactors. It was found that the optimum MLVSS/MLSS ratio should fall within the value of 0.6–0.8 [41].

The interactions of MLSS towards membrane fouling remained contradicted to date. Some researchers noticed that higher MLSS concentration was good for membrane fouling control [35]. However, some studies concluded that higher MLSS concentration would result in lower membrane filterability. This may be because high MLSS concentration would retain particle flocs in the EPS matrix which affects the membrane filtration process [34–43]. Rosenberger et al. [44] found that by increasing MLSS concentration more than 6 g/L helped to reduce membrane fouling but when

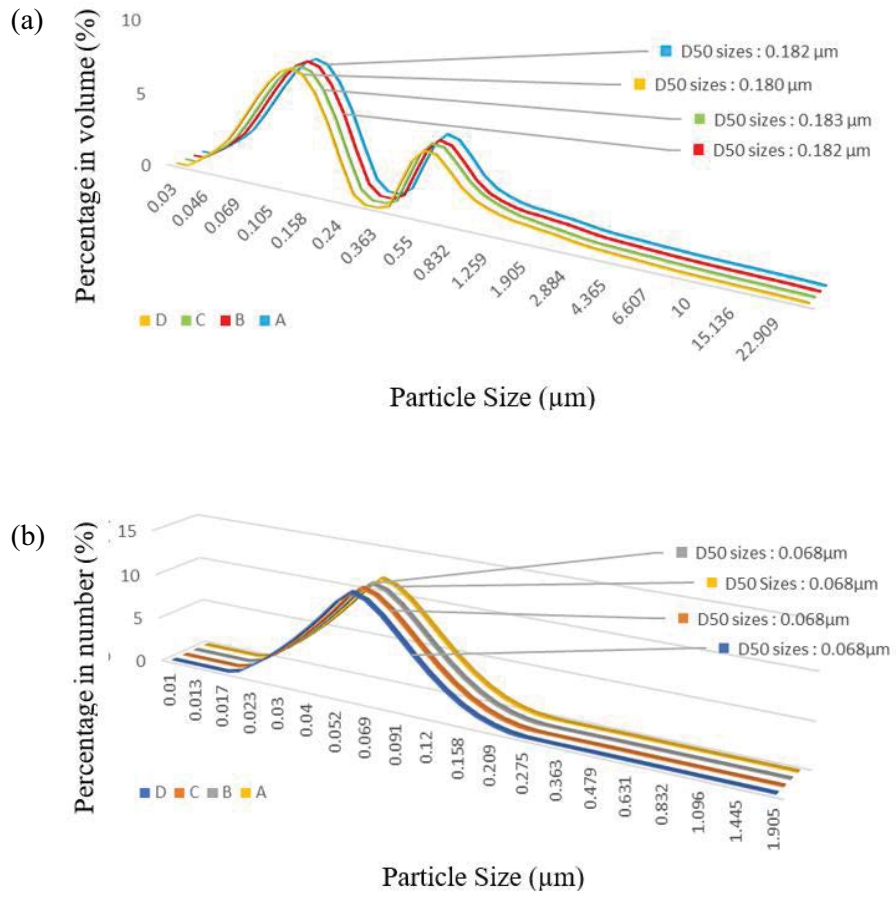

Fig. 7. Sludge floc size distributions based on (a) volume and (b) number counts of different An-MBRs.

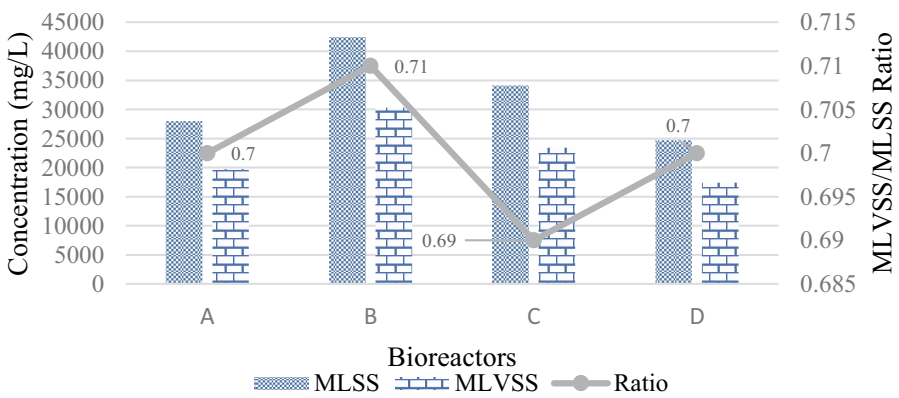

Fig. 8. MLSS and MLVSS concentrations ratio of different An-MBRs.

the MLSS concentration increased more than 15 g/L, membrane would suffer serious fouling. However, Lübbecke et al. [45] found that only above 30 g/L of MLSS, obvious membrane fouling could be observed. On the other hand, Lee et al. [46] observed that low MLSS concentration accelerated the process of having severe membrane fouling. Due to the contradictory points of view from various research studies, it can be deduced that MLSS alone is a weak indicator to explain membrane fouling phenomenon. To know better the main factors causing serious membrane fouling, investigating other biomass characteristics are also require; d [35]. From Fig. 8, bioreactor B shows that higher MLSS concentration contributed to a higher membrane fouling and the bioreactor D with the lowest MLSS concentration was one of the best performers in membrane fouling control. As above-mentioned, MLSS is not the sole membrane fouling indicator. Bioreactor C with relatively higher MLSS did not contribute to higher membrane fouling.

Good performance of the bioreactor D in membrane fouling control may be merely attributed to its lower MLSS concentration as per Figs. 4, 8 and 9. Based on the results obtained from this study, it was found that the main parameters affecting the performance of An-MBRs in membrane fouling control are (i) MLSS and (ii) EPS concentration. Between these two parameters, it was found that EPS concentration particularly polysaccharide is the dominant foulant compared to the MLSS concentration.

4. Conclusion

Among the four An-MBRs (bioreactors A, B, C, and D), it was found that the bioreactor C added with microbe activator with dilution factor of 1/500 showed the best results in terms of COD removal efficiency, EPS concentration removal efficiency (mainly in terms of polysaccharides removal efficiency) and membrane fouling control. POME was used in this research work as the fed in solution which contains COD concentration that is up to 6,000 ± 300 mg/L which is relatively higher compared to domestic wastewater (600–900 mg/L). This can explain why some MBRs performed better in terms of COD removal rate. In this research work, the removal rate of the COD of POME after being treated by the bioreactor C and the membrane filtration is 70.60% ± 2.20% and 79.47% ± 3.74%, respectively. The good result of the bioreactor C may be due to the presence of appropriate amounts of light metal cations in the microbe activator added into it. Light metal cations such as magnesium, calcium, potassium and sodium had been reported able to aid in the anaerobic digestion process, by supplementing nutrients to the bacteria species. Higher than necessary concentration of light metal cations however, exhibited inhibitory properties to the bacteria. Therefore, the metal cations need to be present in suitable concentration such that its presence will not exert pressure to the bacteria species. In this study, except the dilution ratio of 1/500, other dilution ratios did not help the An-MBRs to perform better in terms of pollutants removal rate and membrane fouling control. This could be because the different species of light metal cations will be balanced out against each other in an effect named as an antagonistic effect. Hence, excessive low or high concentration of metal cations is not helpful on the bacteria species in improving An-MBR performance.

The results showed that the membrane fouling had been observed to be in the lowest when the EPS removal rate from the bioreactor is in the highest. This is in consensus with various studies which had identified EPS as the main membrane foulant. Among the protein and polysaccharides, the latter had been recognized to play the main part towards membrane fouling propensity due to its hydrophobic and gelling properties. For the MLSS/MLVSS ratio, the results are indecisive. However, it was found that MLSS concentration increased if the concentration of microbe activator added into the bioreactor increased but it was also noticed that MLSS concentration affected the membrane fouling control performance of An-MBR. An-MBR with higher MLSS concentration tended to foul more seriously. It can be concluded from this study that EPS and MLSS concentrations are the two major parameters affecting the performance of An-MBR. Nevertheless, it was found that EPS, especially polysaccharides concentration plays a more dominant role compared to MLSS concentration in controlling membrane fouling.

Acknowledgements

We would like to extend our gratitude to Universiti Tunku Abdul Rahman for UTAR RESEARCH FUND with project No. IPSR/RMC/UTARRF/2015-C2/N02 and Best

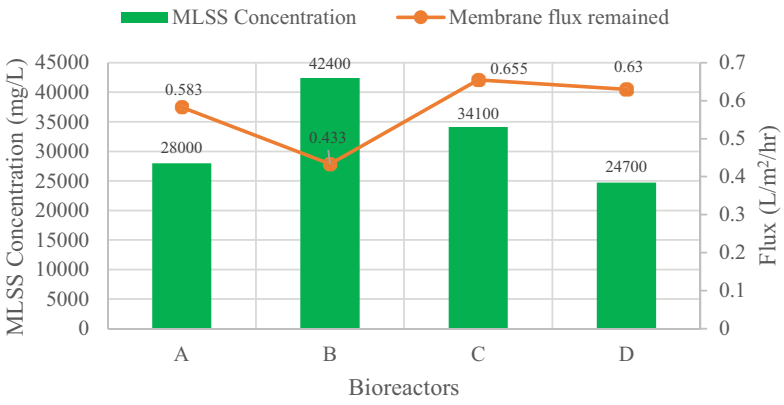

Fig. 9. Effect of different MLSS concentrations on membrane fouling control.

Environmental Technologies (M) Sdn. Bhd. for the fund with vote No. 8062/001.

References

[1] F. Zanetti, G. De Luca, R. Sacchetti, Performance of a full-scale membrane bioreactor system in treating municipal wastewater for reuse purposes, Bioresour. Technol., 101 (2010) 3768–3771.

[2] K. Xiao, S. Liang, X. Wang, C. Chen, X. Huang, Current state and challenges of full-scale membrane bioreactor applications: a critical review, Bioresour. Technol., 271 (2019) 473–481.

[3] J. Sun, P. Liang, X. Yan, K. Zuo, K. Xiao, J. Xia, Y. Qiu, Q. Wu, S. Wu, X. Huang, M. Qi, X. Wen, Reducing aeration energy consumption in a large-scale membrane bioreactor: process simulation and engineering application, Water Res., 93 (2016) 205–213.

[4] S. Vinardell, S. Astals, M. Peces, M.A. Cardete, I. Fernández, J. Mata-Alvarez, J. Dosta, Advances in anaerobic membrane bioreactor technology for municipal wastewater treatment: a 2020 updated review, Renewable Sustainable Energy Rev., 130 (2020) 109936, doi: 10.1016/j.rser.2020.109936.

[5] B.S. Shete, N.P. Shinkar, Comparative study of various treatments for dairy industry wastewater, IOSR J. Eng., 3 (2013) 42–47.

[6] B. Wu, A.G. Fane, Microbial relevant fouling in membrane bioreactors: influencing factors, characterization, and fouling control, Membranes, 2 (2012) 565–584.

[7] H. Lin, M. Zhang, F. Wang, F. Meng, B.-Q. Liao, H. Hong, J. Chen, W. Gao. A critical review of extracellular polymeric subtances (EPSs) in membrane bioreactors: characteristics, roles in membrane fouling and control strategies, J. Membr. Sci., 460 (2014) 110–125.

[8] F. Meng, S. Zhang, Y. Oh, Z. Zhou, H.-S. Shin, S.-R. Chae, Fouling in membrane bioreactors: an updated review, Water Res., 114 (2017) 151–180.

[9] X. Du, Y. Shi, V. Jegatheesan, I.U. Haq, A review on the mechanism, impacts and control methods of membrane fouling in MBR system, Membranes, 10 (2020) 24, doi: 10.3390/membranes10020024.

[10] Best Environmental Technologies, TM Agricultural, 2014.

[11] C. He, C. Yang, S. Yuan, Z. Hu, W. Wang, Effects of sludge retention time on the performance of anaerobic ceramic membrane bioreactor treating high-strength phenol wastewater, Archaea, (2020) 8895321, doi: 10.1155/2020/8895321.

[12] S. Ek, Biological Performance of Palm Oil Mill Effluent (POME) Treatment using An-MBR, Delft University of Technology, Netherlands, 2021.

[13] W.Y. Cheah, P.L. Show, J.C. Juan, J.-S. Chang, T.C. Ling, Microalgae cultivation in palm oil mill effluent (POME) for lipid production and pollutants removal, Energy Convers. Manage., 174 (2018) 430–438.

[14] S.N.H. Abu Bakar, H. Abu Hasan, A.W. Mohammad, S.R.S. Abdullah, R. Ngteni, K.M.M. Yusof, Performance of a laboratory-scale moving bed biofilm reactor (MBBR) and its microbial diversity in palm oil mill effluent (POME) treatment, Process Saf. Environ. Prot., 142 (2020) 325–335.

[15] J. TANG, Treatment of Synthetic Palm Oil Mill Effluent (POME) Using An-MBR: Biological and Filtration Performance, Delft University of Technology, Netherlands, 2021.

[16] W. Chaipetch, W. Khongnakorn, C. Yirong, J. Boonkan, A. Jaiyu, M.J.B. Heran, Performance of a high rate two-stage anaerobic membrane bioreactor (An-MBR) for the treatment of palm oil mill effluent, Bioresources, 17 (2022) 3398–3412.

[17] S.N.B.A. Khadaroo, P. Grassia, D. Gouwanda, P.E. Poh, The impact of thermal pretreatment on various solid–liquid ratios of palm oil mill effluent (POME) for enhanced thermophilic anaerobic digestion performance, J. Cleaner Prod., 261 (2020) 121159, doi: 10.1016/j.jclepro.2020.121159.

[18] M.N. Uddin, M.A. Rahman, J. Taweekun, K. Techato, M. Mofijur, M. Rasul, Enhancement of biogas generation in up-flow sludge blanket (UASB) bioreactor from palm oil mill effluent (POME), Energy Procedia, 160 (2019) 670–676.

[19] F.A.B.M. Lanan, A. Selvarajoo, V. Sethu, S.K. Arumugasamy, Utilisation of natural plant-based fenugreek (*Trigonella foenum-graecum*) coagulant and okra (*Abelmoschus esculentus*) flocculant for palm oil mill effluent (POME) treatment, J. Environ. Chem. Eng., 9 (2021) 104667, doi: 10.1016/j.jece.2020.104667.

[20] N.S.A. Moksin, Y.P. Ong, L.-N. Ho, M.G. Tay, Optimization of photocatalytic fuel cells (PFCs) in the treatment of diluted palm oil mill effluent (POME), J. Water Process Eng., 40 (2021) 101880, doi: 10.1016/j.jwpe.2020.101880.

[21] A.M. Som, A. Yahya, Kinetics and performance study of ultrasonic-assisted membrane anaerobic system using Monod Model for palm oil mill effluent (POME) treatment, Cleaner Eng. Technol., 2 (2021) 100075, doi: 10.1016/j.clet.2021.100075.

[22] Y.Y. Tan, M.M. Bello, A.A. Abdul Raman, Towards cleaner production in palm oil industry: advanced treatment of biologically-treated POME using palm kernel shell-based adsorbent, Cleaner Eng. Technol., 2 (2021) 100079, doi: 10.1016/j.clet.2021.100079.

[23] M.P.O.B. (MPOB), Oil Palm & the Environment, 2014.

[24] M. TmAgri, Best Environmental Technologies, 2014.

[25] J. Barnett, D.E. Richardson, K.R. Stack, T.W.J.A.J. Lewis, Addition of trace metals and vitamins for the optimisation of a pulp and paper mill activated sludge wastewater treatment plant, Appita J., 65 (2012) 237–243.

[26] L.-J. Wu, T. Kobayashi, H. Kuramochi, Y.-Y. Li, K.-Q. Xu, Effects of potassium, magnesium, zinc, and manganese addition on the anaerobic digestion of de-oiled grease trap waste, Arabian J. Sci. Eng., 41 (2016) 2417–2427.

[27] M.S. Romero-Güiza, J. Mata-Alvarez, J.M. Chimenos, S. Astals, The effect of magnesium as activator and inhibitor of anaerobic digestion, Waste Manage., 56 (2016) 137–142.

[28] M. Mellyanawaty, F.M.A. Chusna, H. Sudibyo, N. Nurjanah, W. Budhijanto, Influence of nutrient impregnated into zeolite addition on anaerobic digestion of palm oil mill effluent (POME), IOP Conf. Ser.: Mater. Sci. Eng., 316 (2018) 012069, doi: 10.1088/1757-899X/316/1/012069.

[29] Z. Milán, S. Montalvo, N. Ruiz-Tagle, H. Urrutia, R. Chamy, E. Sánchez, R. Borja, Influence of heavy metal supplementation on specific methanogenic activity and microbial communities detected in batch anaerobic digesters, J. Environ. Sci. Health. Part A Toxic/Hazard. Subst. Environ. Eng., 45 (2010) 1307–1314.

[30] N. Anwar, W. Wang, J. Zhang, Y. Li, C. Chen, G. Liu, R. Zhang, Effect of sodium salt on anaerobic digestion of kitchen waste, Water Sci. Technol., 73 (2016) 1865–1871.

[31] Y. Chen, J.J. Cheng, K.S. Creamer, Inhibition of anaerobic digestion process: a review, Bioresour. Technol., 99 (2008) 4044–4064.

[32] X. Song, W. Luo, J. McDonald, S.J. Khan, F.I. Hai, W. Guo, H.H. Ngo, L.D. Nghiem, Effects of sulphur on the performance of an anaerobic membrane bioreactor: biological stability, trace organic contaminant removal, and membrane fouling, Bioresour. Technol., 250 (2018) 171–177.

[33] A. Anukam, A. Mohammadi, M. Naqvi, K. Granström, A review of the chemistry of anaerobic digestion: methods of accelerating and optimizing process efficiency, Processes, 7 (2019) 504, doi: 10.3390/pr7080504.

[34] F. Delvigne, J.-p. Lecomte, Foam Formation and Control in Bioreactors, M.C. Flickinger, ed., Encyclopedia of Industrial Biotechnology, 2009.

[35] P.K. Gkotsis, A.I. Zouboulis, Biomass characteristics and their effect on membrane bioreactor fouling, Molecules, 24 (2019) 2867, doi: 10.3390/molecules24162867.

[36] J. Liu, Y. Zhao, Y. Fan, H. Yang, Z. Wang, Y. Chen, C.Y. Tang, Dissect the role of particle size through collision-attachment simulations for colloidal fouling of RO/NF membranes, J. Membr. Sci., 638 (2021) 119679, doi: 10.1016/j.memsci.2021.119679.

[37] X. Du, K. Zhang, H. Yang, K. Li, X. Liu, Z. Wang, Q. Zhou, G. Li, H. Liang, The relationship between size-segregated particles migration phenomenon and combined membrane fouling in ultrafiltration processes: the significance of shear stress, J. Taiwan Inst. Chem. Eng., 96 (2019) 45–52.

[38] Y.-J. Liu, D.D. Sun, Particles size-associated membrane fouling in microfiltration of denitrifying granules supernatant, Chem. Eng. J., 181–182 (2012) 494–500.
[39] Y. Liu, X. Li, Y. Yang, W. Ye, S. Ji, J. Ren, Z. Zhou, Analysis of the major particle-size based foulants responsible for ultrafiltration membrane fouling in polluted raw water, Desalination, 347 (2014) 191–198.
[40] W. Yu, D. Zhang, N.J.D. Graham, Membrane fouling by extracellular polymeric substances after ozone pre-treatment: variation of nano-particles size, Water Res., 120 (2017) 146–155.
[41] C.P.L. Grady, G.T. Daigger, H.C. Lim, Biological Wastewater Treatment, Taylor and Francis Group, 1999, p. 1076.
[42] M. Lousada Ferreira, Filterability, Sludge Concentration in Membrane Bioreactors, Delft University of Technology, Netherlands, 2011.
[43] L. Deng, W. Guo, H. Ngo, H. Zhang, J. Wang, J. Li, S. Xia, Y. Wu, Biofouling and control approaches in membrane bioreactors, Bioresour. Technol., 221 (2016) 656–665.
[44] S. Rosenberger, H. Evenblij, S. te Poele, T. Wintgens, C.J.J.o.M.S. Laabs, The importance of liquid phase analyses to understand fouling in membrane assisted activated sludge processes—six case studies of different European research groups, J. Membr. Sci., 263 (2005) 113–126.
[45] S. Lübbecke, A. Vogelpohl, W. Dewjanin, Wastewater treatment in a biological high-performance system with high biomass concentration, Water Res., 29 (1995) 793–802.
[46] J. Lee, W.-Y. Ahn, C.-H. Lee, Comparison of the filtration characteristics between attached and suspended growth microorganisms in submerged membrane bioreactor, Water Res., 35 (2001) 2435–2445.